International Journal of Botany
and Research (IJBR)
ISSN(P): 2277-4815; ISSN (E): 2319-4456
Vol. 7, Issue 2, Apr 2017, 15-20

IMPACT OF HEAVY METALS ON MORPHOLOGICAL RESPONSES OF *ALBIZIA LEBBECK* (L.)

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ABSTRACT

In high anthropogenic pressure locales, pollution of the environment because of heavy metals is a major concern. To understand the impact of these pollutants, this study explores its influence on the morphological features of the important arid legume tree, Albizia lebbeck. This plant was subjected to the differential action of certain heavy metals such as mercury and lead in varying concentrations (1 µM, 10 µM, 100 µM and 1mM). All the different stress treatments had influenced the plant's growth. The least weight of the root was observed at1mM concentration of heavy metal. There was 14.06% and 23.46% reduction in morphological parameters because of mercury and lead, respectively. Increasing heavy metal treatments resulted in decreased germination percentage, shorter root and shoot lengths, and reduced dry mass accumulation in root and shoot. The impact on the root was more than the shoot. In addition, mercury showed more adverse effects than lead; however, the effect was more prominent at 1mM concentration. Nevertheless, different morphological parameters had different effect due to the heavy metals; this differential impact may act as a bio-indicator for pollution by heavy metals.

KEYWORDS: Albizia lebbeck, Mercury, Lead, Heavy Metals & Growth

Original Article

Received: Jan 11, 2017; **Accepted:** Feb 03, 2017; **Published:** Feb 22, 2017; **Paper Id.:** IJBRAPR20173

INTRODUCTION

Heavy metals have an adverse impact environmentally, ecologically and on evolution. The toxicity of the heavy metals is more significant in the localities with high anthropogenic pressure. The chief heavy metals for environmental pollution are cadmium, copper, lead, chromium and mercury. The buildup of heavy metals in the soil has a negative influence on agriculture. The consequences are uncertain food safety and therefore its marketability, stunted crop growth because of phytotoxicity and depletion of soil microorganisms. The metabolic activities of plants modify the geological and biological redistribution of heavy metals. This is usually by the movement of the heavy metals through soil, water and soil. Imperils on the soil by heavy metals need to be continuously monitored as it is known to affect surface water as well as ground water. Mercury and lead are the most toxic, and there are literatures substantiating this fact (Tripathi and Tripathi1998, Gothberg *et al*., 2004, Arunima *et al*., 2015). Mercury is silvery in nature and is found in both organic and inorganic forms. Entry of mercury into the plant is by two main paths: uptake of the oxidized form (Hg (II) or methyl mercury), which is adsorbed on the soil particles, and uptake of the dissolved form in soil water, through roots (Boszke *et al*., 2008). Mercury is reported to have a negative influence even at very low concentrations. Farmers use mercury-based pesticides, which directly influence agricultural plants. Their toxicity effect is borne by photosynthesis and transpiration, which are some of the most fundamental plant physiological processes. The plant's morphology,

anatomy and physiology are imbalanced, as well as pollen germination, pollen tube formation and thereby fruit production (Gill *et al*., 2014). If mercury is the most toxic heavy metal, lead (Pb) is the most widely found heavy metal, according to the environmental protection agency (EPA). Lead is toxic only next to mercury. It enters the food chain because of industries, mining and wrong farming practices, and ultimately causes health issues for humans. Even though Pb is a toxic metal, it is absorbed through the roots and translocated to the shoot. The permissible limit of PB in agricultural soils is 100 mg Pb kg-1 soil. However, this level is on the increase as a result of anthropogenic and industrial actions. This has a negative effect on plant growth, as it modifies the ultrastructure of chloroplast, inactivates the enzymes for CO_2 fixation, photosynthesis, and inhibits lipid peroxidation and antioxidant machinery.

Albizia lebbeck (L.) Benth., locally known as shirish, belongs to the Mimosaceae family. It is a leguminous tree that is commonly found along roadsides. It is also cultivated in industrial areas. This plant is reported to have several medicinal properties. Currently, the spotlight is on plants that can successfully acclimatize the polluted soil and play a significant role in bioremediation. Keeping this trend in mind, this study ventures to understand the consequences of mercury and lead pollution in the soil. The heavy metal toxicity is estimated based on the morphological changes and buildup of proline in *Albizia lebbeck.*

MATERIALS AND METHODS

Albizia lebbeck seeds were collected locally. Only healthy, uniform seeds were used in the investigation. The seeds were surface sterilized to prevent fungal contamination. Diluted sodium hypochlorite was used for this purpose. Thereafter, the seeds were washed repeatedly in distilled water. The seeds were now placed on Whatman filter paper No. 42, which was kept in petri dishes (15cm diameter). This set-up was maintained at room temperature (28°C ± 2) and light intensity of approximately 150 Wm^{-2}. The study was of random type wih five replications of every treatment. Mercury and lead were used in the form of mercuric chloride and lead nitrate, respectively. They were used in the following concentrations: 1µM, 10 µM, 100 µM and 1mM. All solutions were changed on a daily basis, and distilled water served as the control. The plants were subjected to heavy metal stress, and test data were noted on day 4 and day 8. The set-up was maintained for 8 days. Seed germination percentage, root and shoot lengths and dry weight of the root and shoot were the parameters considered in the study. The dry weight was measured after the plant material was kept at 80°C in an oven. Statistical analysis was applied to the data using Duncan's multiple range (DMR) test to drive significance (Duncan, 1955).

RESULTS AND DISCUSSIONS

Environmental pollution by heavy metals is a matter of great concern. Theses metals disturb the ecological, evolutionary, environmental, and nutritional balances. Table 1 illustrates the data of the study on the plant *Albizia lebbeck* L. On heavy metal treatments (mercury and lead at 1 µM, 10 µM, 100 µM and 1mM concentrations), there was reduced seed germination and seedling growth, with the most impact noticed at 1mM concentration. The control plants did not report any significant change in the growth parameters. Mercury showed a 34 % decrease in seed germination at 1mMheavy metal stress, 52% at 100 µM and 64 % at 10 µM. Lead showed a 40 % decrease in seed germination at 1mMheavy metal stress, followed by 58% at 100 µM and 67 % at 10 µM. These data reported the lesser impact of lead compared to mercury, with higher concentrations having more drastic influence on growth. The finding correspond to the ones in literature (Farooqi *et al*., 2009, Pandey and Tripathi 2010). Reduced seed germination is suspected to be because of the rapid breakdown of the reserve food in the seeds subjected to heavy metal stress. A similar impact was noticed in maize

by Kalimuthu & Siva, (1990). They used 20, 50, 100 and 200 µg/ml lead acetate and mercuric chloride, respectively. Iqbal *et al*. (2014) reported a similar influence in tomato plant because of changes in osmotic potential. Jaja and Odoemena (2004) also conducted studies on tomato with ferric and lead salts. They too noticed a shift in osmotic potential. Becerril *et al*., (1989) reported lower water uptake and transport in plants subjected to heavy metals. There may even be embryonic damage and seeding death because of heavy metal stress (Wierzbicka and Obidzinska, 1998). Bewley and Black (1983) blamed the negative impact on the failure in water absorption when treated with heavy metals. Root and shoot lengths were also reduced by heavy metals, with the effect more significant with increasing concentrations. At 1mMconcentration of mercury and lead, 85% and 75% reduced root length and 70% and 60% reduced shoot length were observed, respectively. Root growth was more inhibited than shoot growth, with mercury having a greater influence. Similar observations have been reported in *Albiziasps.*(Farooqi *et al*., 2009, Pandey and Tripathi 2011) and other plants (Jaja and Odoemena 2004, Pandey and Tripathi 2014). Consequently, because of reduction in root and shoot growth, the root and shoot biomass was also reduced by mercury and lead (43% and 54% at 100 µM and 54% and 61% at 1mM, respectively). Here also mercury was more toxic than lead, with higher concentrations having an effect of greater magnitude. *Albizia lebbeck* (Farooqi *et al*., 2009, Iqbal *et al*., 2014), *Albizia procera* [Pandey and Tripathi 2010, 2011], and *Shorea robusta* (Pandey and Tripathi 2014) also reported similar impacts.

Heavy metals have a drastic impact on plant growth, with the magnitude being greater with more concentration. Eun et al. (2000) considered low water potential, lesser nutrient uptake and secondary stress to be the cause in plants subjected to heavy metal stress. The morphology of roots was the most impacted (Oncel *et al.*, 2000, Elloumi *et al.*, 2007, Pant et al., 2011 and Seyyedi, 1999). Heavy metals are also reported to inhibit cell division and cell elongation, as well as reduced mitosis in the meristematic cells of roots and sensitivity of the enzymes in the photosynthetic carbon reduction cycle (De Filippis and Ziegler, 1993). In roots, this results in lesser root cells and surface area for effective water uptake and translocation, which may result in nutrient deficiency. The root accumulates the heavy metals, and limits their transport to the shoots. The effect of Cr, Ni and Hgon *Albizia lebbek* was analyzed by studying its leaf area, root length and shoot length and similar conclusions were drawn (Tripathi and Tripathi, 1999). In laboratory settings, similar findings were reported (Farooqi *et al*., 2009), with lead in the concentrations of 10, 30, 50, 70 and 90 µmol/L producing significant ($p<0.05$) differences in seed germination and seedling length in *A. lebbeck*; 50, 70 and 90 µmol/L lead treatments produced the most severe effect. There are wide-ranging studies on the influence of lead on plant morphology, such as stomata, productivity and yield (Hussain *et al.*,2006), growth tolerance index (Wojas *et al.*, 2007), leaf area (Nosalewicz *et al.*, 2008), inhibition of root elongation (Ghani *et al.,* 2010), plant height (Farooqi *et al.,* 2011) and root, shoot and dry biomass (Azad, 2011). Cell death has been noticed on high lead concentrations (Seregin and Ivanov, 2001). Different treatments of Pb (NO3)2 (200, 400, 600 and 800µM) also exhibited the same influence (Azad, 2011). This study illustrates the fact that heavy metals may inhibit plant growth and development. Because the roots are the most affected plant part, the supply of nutrients and water is impacted. This physiological change indirectly affects hormone production and usage, which may then result in inhibited root and shoot growth. The growth of the plant may become stunted even before the heavy metals are translocated to the shoots.

CONCLUSIONS

This study indicates that both lead and mercury have a negative impact on plant growth. It influences significant growth parameters as well as seed germination. In *A. lebbeck*, mercury had more deleterious effects than lead. Mercury was

highly toxic for seedling growth. The findings of this study indicate the level of tolerance of this species, and its suitability for plantation in heavy metal contaminated areas. The amount of seedling required for effective land coverage is dependent on various factors, such as nature of the heavy metal pollutant in the medium or water for cultivation and concentration. Further investigations need to be performed to appraise the consequences of heavy metals on germination and growth in *A. lebbeck*.

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APPENDIX

Table 1: Seed Germination, Root Length, Shoot Length, Dry Weight (DW) of Root and Dry Weight (DW) of Shoot in Control and Different Treatments of Lead and Mercury in *Albizia lebbeck*, on Day 06 After Induction of Stress

Parameters	Mercury Treatments					Lead Treatments				
	Control	1 µM	10 µM	100 µM	1mM	Control	1 µM	10 µM	100 µM	1mM
Seed Germination (%)	75.92a ±1.2	70.32b ±2.6	64.52c ±3.0	52.50d ±1.2	34.50d ±2.2	75.63a ±2.1	74.72a ±1.9	67.12b ±2.7	58.08c ±2.2	40.08d ±2.9
Root Length (cm plant^{-1})	3.96a ±2.58	3.45b ±2.10 (87.17)	2.83c ±1.98 (71.56)	1.79d ±3.28 (45.42)	0.62e ±2.28 (15.56)	3.90a ±1.66	3.61b ±3.69 (92.57)	3.15c ±1.92 (80.82)	2.15d ±2.25 (55.12)	0.98e ±2.10 (25.12)
Shoot Length (cm plant^{-1})	1.64a ±1.88	1.49b ±1.66 (91.46)	1.31c ±2.00 (79.78)	1.01d ±1.20 (60.90)	0.51e ±1.28 (30.90)	1.69a ±6.64	1.62b ±2.72 (95.76)	1.45c ±1.28 (85.64)	1.11d ±1.44 (66.02)	0.68e ±2.04 (40.21)
DW of Root (g plant^{-1})	0.0070a ±1.01	0.0055b ±0.68 (79.02)	0.0047c ±1.82 (67.90)	0.0031d ±0.48 (43.95)	0.0001e ±1.52 (14.09)	0.0076a ±1.05	0.0066b ±1.26 (86.92)	0.0056c ±2.02 (72.57)	0.0039d ±1.38 (51.43)	0.0018e ±0.48 (23.46)
DW of Shoot (g plant^{-1})	0.0130a ±0.71	0.0116b ±1.12 (89.07)	0.0108c ±0.97 (77.48)	0.0071d ±0.86 (54.96)	0.0036e ±1.28 (28.09)	0.0136a ±0.74	0.0124b ±1.11 (91.50)	0.0112c ±1.15 (82.36)	0.0083d ±2.18 (61.56)	0.0047e ±1.01 (34.56)

The mean values (n=5) in a row followed by different letter for each plant species are significantly different ($P \leq 0.05$) according to Duncan's multiple range (DMR) test. Figures in parentheses represent per cent of control.